Nanosphere

SEP 17 2007

510(k) Summary

This summary of the 510(k) safety and effectiveness information is being submitted in accordance with the requirements of SMDA 1900 and CFR 807.92.

510(k) number k070804

Summary preparation date September 11, 2007

Submitted by

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Contact Sue Kent – Manager, Clinical & Regulatory Affairs

Proprietary names and classifications

For the assay:
Verigene® Warfarin Metabolism Nucleic Acid Test
Regulations: 21 CFR §862.3360 – Drug Metabolizing Enzyme Genotyping System
21 CFR §864.7750 – Prothrombin Time Test
Panels: 91 Toxicology & 81 Hematology
Classification: II
Product codes: ODW – Cytochrome P450 2C9 (CYP450 2C9) Drug Metabolizing Enzyme Genotyping System
ODV – Vitamin K epoxide reductase complex subunit 1 (VKORC1) Genotyping System

For the instrument:
Verigene® System
Regulation: 21 CFR §862.2570 – Instrumentation for Clinical Multiplex Test Systems
Panel: 75 Clinical Chemistry
Classification: II
Product code: NSU - Instrumentation for Clinical Multiplex Test Systems

Common names

For the assays:

warfarin metabolism	*CYP2C9*2*
warfarin panel	*CYP2C9*3*
warfarin	*VKORC1*

For the instrument:
Bench-top molecular diagnostics workstation

Intended uses

- The **Verigene® Warfarin Metabolism Nucleic Acid Test** is an *in vitro* diagnostic for the detection and genotyping of the *2 and *3 alleles of the *CYP2C9* gene and a single-point polymorphism (C to T at position 1173) of the *VKORC1* gene, from EDTA-anticoagulated whole blood samples, as an aid in the identification of patients at risk for increased warfarin sensitivity. The test is intended to be used on the Verigene System.

- The **Verigene® System** is an *in vitro* diagnostic device intended for processing and genotyping multiple genes in a DNA sample utilizing gold nanoparticle probe technology. The Verigene System consists of the Verigene Processor and the Verigene Reader, each with its own onboard proprietary software. The Verigene System is intended to be used by experienced laboratory professionals with training on basic laboratory techniques and on the use of the system components.

Predicate device

Third Wave Technologies, Inc., Invader® UGT1A1 Molecular Assay (k051824)

Device descriptions

The Verigene System is an *in vitro* diagnostic device for processing and genotyping multiple genes in a DNA sample. The Verigene System consists of two instruments, the Verigene Processor and the Verigene Reader, and utilizes single-use, disposable Test Cartridges to process and genotype multiple genes in a DNA sample in approximately 1½ hours.

The analysis sequence is the same for each of the three tests (i.e., *CYP2C9*2 and *3 and VKORC1*). After extracted and purified DNA, mixed with hybridization buffer, is loaded into the sample well of the Test Cartridge, it is ready for processing and is inserted into the Verigene Processor. An internal barcode reader reads the cartridge ID and sends the information to the Verigene Reader. From this information, the Verigene Reader establishes the hybridization parameters and starts the hybridization process.

The genotyping process occurs with a hybridization of the target analyte to a synthetic gene-specific oligonucleotide capture strand on the Test Cartridge's substrate. A synthetic mediator target-specific oligonucleotide is included with the test-specific sample buffer to form a hybridization "sandwich" with the gene sequence of interest. Washing steps following the target hybridization remove the unbound DNA from the hybridization chamber. A probe, composed of a gold nanoparticle with covalently bound oligonucleotides complementary to a sequence on the intermediate oligonucleotide, is introduced after the target wash. After the probe hybridization is completed, a series of washing steps remove the unbound probe from the hybridization chamber. A two-part signal enhancement reagent is added to the hybridization chamber and reacts with the gold nanoparticle to amplify the signal for the Verigene Reader scanning and analysis.

Upon completion of the genotyping process, the user removes the Test Cartridge from the Verigene Processor which is now ready for the next test.

Once the reagent portion of the Test Cartridge is removed by the user, the substrate is inserted into the Verigene Reader. The Verigene Reader illuminates the signal-enhanced nanoparticles specifically bound to either the wild type or mutant captures for the gene. A photosensor reads the relative brightness of each spot and the Verigene Reader outputs a result based on relative levels of brightness of the wild type to mutant signals.

Comparison to technological features of the predicate device

The following is a comparison of the Nanosphere, Inc., Verigene System to the Third Wave Technologies, Inc., Invader® UGT1A1 Molecular Assay:

Characteristic	Third Wave Technologies, Inc., Invader® UGT1A1 Molecular Assay	Nanosphere, Inc., Verigene® Warfarin Metabolism Nucleic Acid Test
DNA sequence detection	Detects specific DNA sequences through direct recognition of DNA targets	Same as predicate
Reaction conditions	1) No thermal cycling – isothermic reaction 2) Utilizes signal amplification 3) Reactions occur in multiple plastic microtiter wells	1) Same as predicate 2) Same as predicate 3) Reactions occur on a single glass slide using microfluidics, with up to 32 cartridges (slides) hybridized and enhanced simultaneously
Assay results	Assay signal results are interpreted by a software program and are assigned a genotype that is presented to the end-user in a report format.	Same as predicate

Performance characteristics

Reproducibility –

In an initial reproducibility study at each of three sites, five genomic DNA samples, covering all possible genotypes for all three alleles, were each tested in triplicate on a daily basis by the same operator for three days. One site performed the same reproducibility testing twice each day, using two different operators. The table below shows the number of samples tested, correct calls, incorrect calls, no calls, and percent agreement (or call rate) by locus for each of the three sites.

Initial reproducibility results

Site	Locus	Samples Tested	Correct Calls	Incorrect Calls (Mis-Calls)	No Calls	Call Rate (% agreement)
Site 1	*2C9*2*	90	85	0	5*	94%
	*2C9*3*	90	85	0	5*	94%
	VKORC1	90	85	0	5*	94%
Site 2	*2C9*2*	45	40	0	5*	89%
	*2C9*3*	45	40	0	5*	89%
	VKORC1	45	40	0	5*	89%
Site 3	*2C9*2*	45	41	0	4	91%
	*2C9*3*	45	41	0	4	91%
	VKORC1	45	41	0	4	91%

*includes 1 pre-insertion error

In a second reproducibility study (see table below) that evaluated three common DNA extraction methods, aliquots of a panel of 23 blood specimens were utilized. At each of the 3 sites, 1 operator extracted the DNA from each of the 23 aliquots of blood and ran the assay. Each site used a different DNA extraction procedure/kit. 3 lots of cartridges were tested. 1 re-test run was performed if there was a "no call" or "mis-call" on the first run but no mis-calls were observed. The genotypes of the DNA samples were confirmed by bi-directional sequencing. The genotypes of the 23 samples included:

- *2C9*2*: 18 wild type, 4 heterozygous, 1 mutant
- *2C9*3*: 20 wild type, 3 heterozygous, 0 mutant
- *VKORC1*: 9 wild type, 11 heterozygous, 3 mutant

Extraction method reproducibility results

Site	Samples Tested	Run	Genotyping Calls Made	Correct Calls	Incorrect Calls (Mis-Calls)	Call Rate (% agreement)
Site 1	23	After run 1	21	21	0	91%
		After run 2	23	23	0	100%
Site 2	23	After run 1	21	21	0	91%
		After run 2	23	23	0	100%
Site 3	23	After run 1	22	22	0	96%
		After run 2	23	23	0	100 %

Accuracy (percent agreement compared to bi-directional DNA sequencing) –

A total of 238 samples were analyzed using the Verigene Warfarin Metabolism Nucleic Acid Test at three sites and by bi-directional sequencing analysis at an independent reference laboratory (see results in the tables below). All purified DNA samples were from whole blood collected using EDTA as the anticoagulant. These data are based on the original run only (i.e., no re-testing was performed). Three Verigene cartridges were defective and failed to run due to pre-insertion errors.

CYP2C9*2 **method comparison results**

Sequence analysis		Wild-type (wt)	Heterozygous (het)	Mutant (mut)	Incorrect Call Rate (Mis-Calls)	Correct Call Rate [% agreement] (No Calls)
	wt	176	0	0	0% (0)	92% (15)
	het	0	35	0	0% (0)	87% (5)
	mut	0	0	2	0% (0)	67% (1)

CYP2C9*3 **method comparison results**

Sequence analysis		Wild-type (wt)	Heterozygous (het)	Mutant (mut)	Incorrect Call Rate (Mis-Calls)	Correct Call Rate [% agreement] (No Calls)
	wt	182	0	0	0% (0)	91% (17)
	het	0	30	0	0% (0)	88% (4)
	mut	0	0	1	0% (0)	100% (0)

VKORC1 1173 **method comparison results**

Sequence analysis		Wild-type (wt)	Heterozygous (het)	Mutant (mut)	Incorrect Call Rate (Mis-Calls)	Correct Call Rate [% agreement] (No Calls)
	wt	79	0	0	0% (0)	91% (8)
	het	0	97	0	0% (0)	89% (12)
	mut	0	0	34	0% (0)	97% (1)

The Verigene Warfarin Metabolism Nucleic Acid Test results will be reported only if calls are made for all three targets (i.e., the panel must be complete, based on calls made for *CYP2C9*2, CYP2C9*3,* and *VKORC1* calls). If only one or two allele calls are made, the entire test's results are not reported on the screen or printout.

Therefore, from an overall panel view, the total panel read rate was 91.1% (=214/235).

Limit of Detection (analytical sensitivity)–

The table below shows the results of a DNA concentration study. When DNA concentrations outside of the range of 40 ng/µL – 400 ng/µL are studied, no mis-calls are made but the call rate decreases. At 40 ng/µL, the call rate is 92%.

Limit of detection study results

DNA Concentration	Number of Cartridges	Correct Calls	Incorrect Calls (Mis-Calls)	No Calls	Call Rate (% agreement)
30 ng/µL	12	9	0	3	75%
40 ng/µL	12	11	0	1	92%
200 ng/µL	12	12	0	0	100%
400 ng/µL	12	12	0	0	100%
500 ng/µL	12	11	0	1	92%

Other characteristics of the Verigene Warfarin Metabolism Nucleic Acid Test

Characteristic	*CYP2C9*2*	*CYP2C9*3*	*VKORC1*
Interferences	Performance not affected by • magnetic beads • heparin • hemoglobin • magnesium chloride • lithium chloride • other possible interferences are not known.		
Reagent stability	• The Test Cartridges are to be stored from 2°C to 8°C until the expiration date printed on the label. • The Sample Buffer is to be stored from 2°C to 8°C until the expiration date printed on the label. • Neither the Test Cartridges nor the Sample Buffer should be frozen.		
Precautions and warnings	In the *CYP2C9* gene, additional rare mutations other than R149C (*CYP2C9*2*) and I359L (*CYP2C9*3*) have been observed. These rare CYP2C9 alleles are not detected by this test. NOTE: The prevalence of these additional alleles is low and there is insufficient information in the scientific literature to predict the impact this polymorphism will have on an individual's sensitivity to warfarin.		In the *VKORC1* gene, additional rare polymorphisms other than 1173C>T have been observed. These rare VKORC1 alleles are not detected by this test.

Conclusion

The above pre-clinical and clinical test results support the safety and effectiveness of the devices – the Verigene System and Verigene Warfarin Metabolism Nucleic Acid Test.

DEPARTMENT OF HEALTH & HUMAN SERVICES

Public Health Service

Food and Drug Administration
2098 Gaither Road
Rockville MD 20850

Nanosphere, Inc
c/o Ms. Sue Kent
4088 Commercial Avenue
Northbrook, IL 60062

SEP 17 2007

Re: k070804
Trade/Device Name: Verigene Warfarin Metabolism Nucleic Acid Test, Verigene System
Regulation Number: 21 CFR 862.3360
Regulation Name: Drug metabolizing enzyme genotyping system
Regulatory Class: Class II
Product Code: ODW, ODV, NSU
Dated: August 7, 2007
Received: August 8, 2007

Dear Ms. Kent:

We have reviewed your Section 510(k) premarket notification of intent to market the device referenced above and have determined the device is substantially equivalent (for the indications for use stated in the enclosure) to legally marketed predicate devices marketed in interstate commerce prior to May 28, 1976, the enactment date of the Medical Device Amendments, or to devices that have been reclassified in accordance with the provisions of the Federal Food, Drug, and Cosmetic Act (Act) that do not require approval of a premarket approval application (PMA). You may, therefore, market the device, subject to the general controls provisions of the Act. The general controls provisions of the Act include requirements for annual registration, listing of devices, good manufacturing practice, labeling, and prohibitions against misbranding and adulteration.

If your device is classified (see above) into either class II (Special Controls) or class III (PMA), it may be subject to such additional controls. Existing major regulations affecting your device can be found in Title 21, Code of Federal Regulations (CFR), Parts 800 to 895. In addition, FDA may publish further announcements concerning your device in the Federal Register.

Please be advised that FDA's issuance of a substantial equivalence determination does not mean that FDA has made a determination that your device complies with other requirements of the Act or any Federal statutes and regulations administered by other Federal agencies. You must comply with all the Act's requirements, including, but not limited to: registration and listing (21 CFR Part 807); labeling (21 CFR Parts 801 and 809); and good manufacturing practice requirements as set forth in the quality systems (QS) regulation (21 CFR Part 820).

This letter will allow you to begin marketing your device as described in your Section 510(k) premarket notification. The FDA finding of substantial equivalence of your device to a legally marketed predicate device results in a classification for your device and thus, permits your device to proceed to the market.

If you desire specific information about the application of labeling requirements to your device, or questions on the promotion and advertising of your device, please contact the Office of In Vitro Diagnostic Device Evaluation and Safety at (240) 276-0490. Also, please note the regulation entitled, "Misbranding by reference to premarket notification" (21CFR Part 807.97). You may obtain other general information on your responsibilities under the Act from the Division of Small Manufacturers, International and Consumer Assistance at its toll-free number (800) 638-2041 or (240) 276-3150 or at its Internet address at http://www.fda.gov/cdrh/industry/support/index.html.

Sincerely yours,

Jean M. Cooper, M.S., D.V.M.

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Director
Division of Chemistry and Toxicology
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Enclosure

Indications for use

510(k) Number (if known): K070804

Device Name: Verigene® System

Indications for Use: The Verigene System is an *in vitro* diagnostic device intended for processing and genotyping multiple genes in a DNA sample utilizing gold nanoparticle probe technology. The Verigene System consists of the Verigene Processor and the Verigene Reader, each with its own onboard proprietary software.

Device Name: Verigene Warfarin Metabolism Nucleic Acid Test

Indications for Use: The Verigene Warfarin Metabolism Nucleic Acid Test is an *in vitro* diagnostic for the detection and genotyping of the *2 and *3 alleles of the *CYP2C9* gene and a single-point polymorphism (C to T at position 1173) of the *VKORC1* gene, from EDTA-anticoagulated whole blood samples, as an aid in the identification of patients at risk for increased warfarin sensitivity. The test is intended to be used on the Verigene System.

Prescription Use __X__ and/or Over-The-Counter Use _____
(Part 21 CFR 801 Subpart D) (21 CFR 801 Subpart C)

(PLEASE DO NOT WRITE BELOW THIS LINE-CONTINUE ON ANOTHER PAGE IF NEEDED)

Concurrence of CDRH, Office of In Vitro Diagnostic Devices (OIVD)

Carol C. Benson

...ion Sign-Off

Office of In Vitro Diagnostic Device
Evaluation and Safety

K070804